GENERAL MANAGEMENT ASSISTANCE CONTRACT (GMAC)

Contract No: 674-C-00-01-00051-00

"Sceletium Tortuosum in rural SA"

Grant number: 0126-1003-G-GA34

AFRIPLEX

This report was produced for review by the USAID. It was prepared as a performance milestone under Mega-Tech, Inc.'s prime contract. The contents of this report address activities performed under USAID/South Africa's Strategic Objective No. 9: Increased Market-Driven Employment Opportunities

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Activity Summary and achievements:

The main focus of this project was research regarding propagations and cultivation of Sceletium tortuosum, and the cost effectiveness of different cultivation techniques before it could be implemented at farm level. Reproductive and vegetative plant material from Sceletium tortuosum was collected from the wild. Selection took place on the grounds of mesembrine content as well as yield per plan. Research regarding different propagation techniques was undertaken (seed germination, cuttings and tissue culture). Comparisons between efficiency of reproduction, time taken and costs involved were made (year 1 and 2). The economically most viable method was implemented at small-scale grower level (3rd year).
Simultaneously with the propagation studies, field trials were implemented at Elsenburg to focus on appropriate cultivation methods (planting distance; light water and nutrient requirements). Attention was given to harvesting (sustainable and total harvest) and post-harvesting handling. An economic comparison was drawn up to determine the better practice between replacing after each harvests of sustainable harvesting of plants, keeping them in the soil for successive seasons. The nature of the plant makes drying quite challenging, without loosing too much of the mesembrine content. This was investigated at a level possible to implement at small-scale grower level, with preference given to female growers. Product harvested from the trails plots (year 1 and 2) were sent as samples to different companies, locally and overseas, dealing in medicinal plants in order to secure markets for the product once it was implemented in small-scale farming activities.

The attached Grant Activity Completion Report presents the program and its achievements in more detail.

Contents of this report:

1. Grant Activity Completion Report (February 2006)

2. Annual Report (September 2005)

Grant Activity Completion Report

1. Name of Organisation	Afriplex
2. Grant Activity	Sceletium tortuosum in rural South Africa

3. Briefly describe the grant objectives achievements and impact as a result of the grant activities implemented during the grant period.

The objectives of the grant were to look at the cultivation practices of Sceletium tortuosum. In nature the plant grows in the shade of trees, shrubs or rocks. An investigation into the shade and water requirements of Sceletium in cultivation was investigated by means of field and controlled environment trials. The trials were conducted at three different locations to determine the optimum climatic conditions for the crop.

No standard or method for extracting mesembrine (the active ingredient) from Sceletium existed. Another objective was to develop a suitable method for the extraction and quantifying the mesembrine content of the plant.

The project also aimed at micro-propagating the plants in order to have a large number of plants in a small time. The best options of propagation for small-scale farmers were investigated.

Lastly the aim was to determine, by means of DNA fingerprinting, plants with high mesembrine content for commercialization.

4. Briefly discuss the implementation process, including lessons learned and recommendations

Plants were germinated from seed (and the germination protocols recorded) and transplanted at three different locations (Osdrif, Swellendam and Wupperthal) and monitored monthly for growth rate. A number of plants were also placed in a protected environment and the amount of water needed for optimum growth was monitored and determined.

About five months after transplanting the plants in the field, the plants at Osdrif started dying and almost overnight 80% of the plantation was dead. Soil samples revealed a soil borne fungus, but not in high enough concentrations to do such damage. None of this was observed at any of the other sites. The only conclusion that was reached was that the climate of Osdrif is too wet and not suitable for the cultivation of Sceletium.

Cuttings were made from Sceletium plants, the success rate and ease of rooting recorded and propagation protocols determined for the making of cuttings. This proved to be the most viable and economic method of propagating Sceletium.

Literature searches revealed methods of extraction of alkaloids from plant material. The initial methods used chloroform and/ or methanol in the extraction process. This was applied and modified to extract mesembrine from Sceletium. Ethanol (which is cheaper and less harmful) is used in the modified method. A method for the extraction of mesembrine from Sceletium was recorded.

Micro propagation of Sceletium showed to be more challenging. The researchers could not get a 'clean' culture to produce the plant in tissue culture, due to the presence of an endophyte (a fungus living in symbiosis with the plant in the cells). The budget and time period for determining the DNA fingerprint of high yielding plants was not enough to get to a conclusive result, but results obtained were reported.

The relative ease of propagation of plant material and the fact that the project was in possession of high yielding mesembrine plants led to the establishment of half a hectare of Sceletium with a disabled group in Calvinia. Calvinia was selected because it is an area where the plants occur naturally. The plants are growing well, the group has harvested the plants and has a greenhouse to multiply their own material, equipment to dry and mill the plant material and the market for the product is increasing.

5. Public Dissemination: GMAC requires that all grant activity deliverable(s) of the grant activity (e.g., a report or survey) must be made available to the general public. Briefly discuss how the grants activities and results were made accessible to interested parties.

All the project activities were recorded and reported on to Megatech. One of the deliverables was a document for small-scale farmers on the propagation, cultivation, and processing of Sceletium. These were made available to the group in Calvinia who received training in that respect. Representatives of Department of Agriculture and Surplus People's Project (SPP) who are overseeing the overall activities the disabled group in Calvinia also attended the training and received the documentation.

Natural Botanicals often receive phone calls from interested farmers or groups who want to cultivate Sceletium. Information gathered during the duration of the project is given to them.

The research results, outsourced to Universities were reported on in the relevant Universities' annual research and output reports.

Marianna Smith gave a presentation in September 2004 at the Southern African New Crop Research Association (SANCRA) titled "Sceletium tortuosum – a natural Prozac?"

Signature of Grantee _____ Date 15/02/06

GMAC Grantee Narrative Report

Grantee AFRIPLEX

Agreement No. 0126-1003-G-GA34

Report for the period:

☐ **October 1, 2004 through September 30, 2005**

Please complete a one-two page narrative summary of your main accomplishments between October 1, 2004 – September 30, 2005. To prevent you from re-inventing the wheel, it is recommended that you work through the two narrative reports that you are required to submit to Mega-Tech for the specified reporting period and capture essential accomplishments related to your milestones.

The section of the project dealing with the cultivation practices and the suitability of various locations came to an end during the reporting period. The research into the most suitable location for cultivation of Sceletium delivered some interesting results. Three locations were selected to establish trial plantations of *Sceletium tortuosum*. The one was on the farm, Osdrif in the Worcester district of the Western Cape. A second plantation was at the farm Nooitgedacht in the Swellendam district and the third at Eselbank, a substation of the Moravian Mission station, Wupperthal in the Cedarberg. The diameter of plants was measured at different intervals and was used as a measure of plant vigor. Fifty percent, or half of the size of the plants were harvested, weighed (for fresh weight determination) and the active ingredient, mesembrine, was determined on this sample. At some of the sites large numbers of the plants died and percentage plants survived at each location was also noted. In conclusion it can be said that *Sceletium tortuosum* can be successfully implemented in plantations at any of the three sites in the Western Cape. The location will not influence plant yield or biomass, but plants have a better chance of survival at Eselbank, then at Nooitgedacht and the worst chance of survival at Osdrif. *Sceletium tortuosum* is grown and marketed for the secondary metabolite, an alkaloid called mesembrine. The industry is starting to set standards and require a certain minimum amount of mesembrine in plant material. Plants at Eselbank produced significantly more mesembrine than plants at Osdrif. In terms of location for plantations to obtain optimum amounts of mesembrine, it will be best to grow plants at Eselbank, with Nooitgedacht as a second choice and Osdrif the least preferred choice. Secondary metabolites are usually produced when plants are under stress. The climate at Osdrif is the mildest of all the sites and that might contribute to the smaller amount of mesembrine present in plants grown at Osdrif. Harsh climate, with very cold winters and extremely hot summers are experienced at Eselbank, and might be the reason for the higher mesembrine determined in plants grown there.

The experimental plot at Osdrif (near Worcester in the Western Cape) was planned to give a whole lot of information on the cultivation practices of *Sceletium tortuosum*. Planting distance (amount of plants per square meter), percentage shade (40, 20 and 0%) and the optimum amount of plant material harvested from the plant (25, 50 or

75%) were to be determined. In nature the plant always occurs in the shade of another plant, small bush or rock, which gives a perception that the plant needs a shade requirement for survival. Another task was also to look at the optimum time of harvest (month of the year) during the experimental time. According to history, and Van der Stel's diaries since 1685, the indigenous people harvested the plant in October. The experimental layout wanted to verify or disprove this. The experimental layout also allowed for investigating the influence of different types of mulch (no mulch, black plastic or bark) on plant biomass or yield. Mulch is used to control weeds and keep more moisture in the soil. Less competition and more soil moisture available to the plant might influence the yield.

The conclusion drawn from the experimental results indicated that different levels of shade did not influence the survival rate of *Sceletium* plants at Osdrif. However, at this point it is noteworthy to remark that at Helpmekaar Trust in Calvinia, where a semi-commercial plantation was established and where *Sceletium* occurs naturally, NO plants survived in the full sun and few mortalities (less than 2%) were experienced under 40% shade. The climate at Osdrif is much less harsh than in Calvinia and might be the reason for detecting no difference between the survival rate of plants at different levels of shade at the Osdrif locality. Mesembrine content in the plants tested did not show any significant difference in plants grown under different conditions of shade (0, 20 or 40% shade). Mesembrine content shows a definite peak between 216 and 224 days after establishment. This is from late October to the beginning of November. The indigenous people harvested *Sceletium* during October. They gathered the plants, fermented it in sheepskin bags, dried it in the sun and stored their supply for the rest of the year. Our results confirmed that the mesembrine content is higher during late October and early November. The growing season for *Sceletium* is during the winter. In early summer the rainfall starts to decline, temperatures increase and the plant starts to experience more stress. When plants are under stress they produce more secondary metabolites, i.e. more mesembrine.

No big differences in the diameter of plants grown with different soil mulches were measured. Plants grown with bark as mulch produced the highest biomass, with black plastic the second highest biomass was experienced and no mulch produced the lowest biomass. It was expected that soil mulch will be beneficial in production of biomass as it keeps weeds under control and keeps moisture much longer in the soil.

A second experiment to look into the cultivation practices of *Sceletium tortuosum* was conducted at Elsenburg, under controlled conditions. The optimum amount of water for the growth of *Sceletium* was investigated. The plants were kept in a greenhouse, where rain would not influence the amount of water plants receive. Single plants were transplanted in 10 liter and 20 liter pots respectively. Potted plants received a total of 250 ml, 500 ml or 1000 ml weekly. These amounts were applied once a day (seven times per week), three times a week or once per week. There were four replicates per treatment per pot size. Plants receiving water once per week and potted in the smaller pots (10 liter) had higher yields than plants in the 20 liter pots. More soil is present in the bigger pots, and although the pots received the same amount of water, less water was available for the plant in the bigger pots, hence the lower yield in the bigger pots.

The same phenomenon was observed with potted plants receiving water 3 times per week. A plant growing in a 10 liter pot, receiving 1000 ml of water had the highest

yield, and plants with the same water regime in 20 liter pots the lowest yield. This indicates that a little water at intervals is more beneficial to plant growth and yield.

The most interesting observation is where plants are watered every day of the week. The plants growing in 20 liter pots, receiving a total of 1000 ml of water per week had the highest total yield of all the water regimes tested. Of interest here is to note that the corresponding set of plants, but grown in 10 liter pots had a low yield, even lower than the potted plants who received either 250 or 500 ml of water per week. The plants in the 10 liter pots, receiving a total of 1000 ml per week, but applied in small amounts every day was too wet for the plant and the yield went down.

In conclusion it can be said that *Sceletium* plants have higher yields when small amounts of water are applied at regular intervals, but great care has to be taken to ensure the soil surrounding the plants do not become waterlogged.

A semi-commercial plantation of 0.5ha (20 000 plants) was established at Helpmekaar Trust near Calvinia. The plantation is doing extremely well and the beneficiaries of the Trust already harvested 6.5 kilograms of dried *Sceletium* (1kg dry = 20 kg wet material). The dried plant material was sold at a price of R1000 per kilogram.

In my capacity as the grantee agent, I hereby certify that to the best of my knowledge, the above information is accurate.

Authorised Signature: _____

Date: _____

GMAC Grantee Counterpart Funding

Grantee AFRIPLEX

Agreement No. 0126-1003-G-GA34

Report for the period:

☐ **October 1, 2004 through September 30, 2005**

NGO and Third Party Counterpart Contribution Tracking

Source	Cash Contributions (ZAR)		Rand Value of In-Kind Contributions		Comments
	Planned[1]	Actual[2]	Planned[3]	Actual[4]	
NGO - Afriplex	0	0	R29,717	R29,717	Accounting, marketing and product development of Sceletium
Farmers	0	0	R6,240	R12,000	Use of labour and farm infrastructure, amounted to more, because a semi-commercial plantation was established.
Natural Botanicals	0	0	R100,926	R118,902.34	Salaries, benefits, travel and per diem. Actual values are more, because more time was spent on the semi-commercial plantation.

In my capacity as the grantee agent, I hereby certify that to the best of my knowledge, the above information is accurate.

Authorised Signature: _____

Date: _____

[1] Planned Cash Contributions refers to your grant agreement amount for the period Oct 1, 2004 – September 30, 2005

[2] Actual Cash Contributions refers to your grant agreement amount for the period Oct 1, 2004 – September 30, 2005

[3] Planned In-kind Contributions refers to your grant agreement amount for the period Oct 1, 2004 – September 30, 2005

[4] Planned In-kind Contributions refers to your grant agreement amount for the period Oct 1, 2004 – September 30, 2005